OBSERVATION ON LIVING SPECIMENS OF IRIDIA DIAPHANA, A SPECIES OF FORAMINIFERA.

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While at the Tortugas laboratory of the Carnegie Institution of Washington, through the kindness of the director, Dr. Alfred G. Mayor, I collected numerous foraminifera. Among these were living specimens which are apparently identical with or close to the species described by Heron-Allen and Earland as *Iridia diaphana*¹.

Specimens are attached to various objects but the most accessible are those which are adherent to the broad leaves of *Posidonia* which covers the bottom in shallow water in various places.

In general the test of the Tortugas specimens of Iridia is made largely of calcareous sand grains (pl. 19, fig. 1) in a slightly raised dome, but larger specimens usually are more or less irregular in outline. The upper surface only is composed of acclutinated material. the lower surface and the lining of the upper side being of thin chitinous material forming an enclosing membrane. The calcareous material may be entirely dissolved in weak acid leaving this chitinous envelope intact. In its living state viewed from above the interior of the test is not visible, due to the opaque wall of sand grains. With a thin scalpel the entire test may easily be removed from the leaf of Posidonia and transferred to the water. If placed upside down and examined with low power more of the structure of the test can be seen. A small sector of this is shown in plate 19, figure 2. Seen from below there is at once distinguished a central more or less homogeneous mass of a light vellowish brown color surrounded by a slightly darker periphery. Outside this is a peripheral band to the edge of the test, of lighter color with practically no protoplasm in most of the area, but here and there at intervals with irregular bands passing from the central protoplasmic mass to the periphery of the test. These are of a light yellow color in general like that of the central mass. In each of these radial bands appear several more or less distinct channels and from these the protoplasm streams back and

¹ Trans. Zool. Soc. London, vol. 20, 1914, p. 371, pl. 36.

PROCEEDINGS U. S. NATIONAL MUSEUM, VOL. 57-No. 2308.

forth from the exterior. The only connection between the central cavity of the test and the exterior seems to be through these radial channels. When placed with the convex side downward the chitinous layer may be gently pressed, causing the protoplasmic mass of the central portion to sway back and forth; but it is held firmly within the chitinous envelope unless this is ruptured by the rough treatment. The chitinous layer is, however, tough and capable of considerable resistance.

By reflected light the protoplasmic portion is of a peculiar light yellowish brown color against the white of the sand grains of the test with the lines of the radial channels and the peripheral portion of the central mass of a deeper orange color. With high power this deeper color is seen to be due to the presence of great numbers of unicellular algae, which seem to be rather closely confined to these areas or at least to be concentrated there in greater numbers. These algae are brownish in color, of very uniform size, about $6-7 \mu$ in length and apparently have a commensal relation with the protozoan as will be noted later.

Usually within five minutes after the foraminifer has been detached from the Posidonia and placed in fresh sea water pseudopodia appear at the periphery. These are thrust out rapidly and carry with them numbers of the unicellular algae already mentioned. These seemingly adhere to the sides of the fine pseudopodia, as shown in plate 19, figure 3. The pseudopodia themselves are largely of clear protoplasm with fine "knots" at irregular intervals of slightly more compact material. There is a tendency for the algae to collect at the junctions of the pseudopodia. In the finer pseudopodia there is often a movement in the opposite directions at the same time, one side carrying the "knots" and algae outward, the other side carrying them backward toward the test. This movement may be suddenly interrupted and all flow in one way or the other or each become reversed. When steadily moving the protoplasm of the finer pseudopodia is carried along at an average speed of about 1 mm. a minute from actual measuments with micrometer scale and stop watch.

The unicellular algae evidently have a commensal relation to the foraminifer, as they are usually associated with the pseudopodia. When the pseudopodia are extended and active nearly all the algae are outside the actual test and when quiescent they are apparently stored in the area at the peripheral portion of the central mass and in the radial channels, as already noted.

The protoplasmic body of the animal has the power of leaving the test at will. This was noted in cases where specimens were detached and left over night in sea water and in the morning found at some distance from their empty test. Whether this withdrawal was through the radial canals or through some breaking down of the chitinous

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lining could not be determined. These specimens had practically no included algae. When such specimens have left their test they are capable of extended and fairly rapid movement. One such specimen which had left its test voluntarily is shown in plate 20, figure 1. It is elongate, with a definite "head" to the body and numerous pseudopodia streaming behind, as well as a few in front. The "body" portion with its pseudopodia formed a line more than an inch in length. During its movement records of its rate were taken and these were from 1 minute and 40 seconds to 2 minutes for a distance of a millimeter. Later other specimens were found to have a much more rapid rate of motion as will be given.

The movements and changes in this specimen seem worthy of note. After traveling for a considerable distance in the elongate form (pl. 20, fig. 1) it became concentrated into a more rounded form, which semingly was about to divide into two portions by its constrictions, but later started out again, in an elongated form. These (pl. 20, figs. 2-6; pl. 21, figs. 1-7) were drawn with camera lucida at intervals while the whole protoplasmic mass was in slow but rather steady motion. Toward the end of this series motion became less rapid and nearly ceased, but later a clearer "tongue" of protoplasm was thrust out (pl. 21, fig. 7) and motion again became became more rapid.

After the elongate form and more rapid movement had been resumed portions were cut off with a scalpel. Where a small portion of the end was severed, the detached mass quickly became globular by concentration of the protoplasmic portions; then after a short time numerous fine pseudopodia were sent off very rapidly in all directions until fusion took place with the original mass, which also during the same period had sent back numerous fine pseudopodia from the direction of the cut. When fusion of the pseudopodia takes place the smaller severed portion sets up a rapid streaming toward the larger mass until the two are once more united, when shape and motion are carried out as before.

Where a small mass was removed and placed at some distance from the larger mass the same process was repeated, but after fusion was not made, owing to the distance, the smaller mass became concentrated in a subspherical mass, where it remained quiescent for two days, then gradually broke down and disintegrated. This smaller portion probably had no nuclear material and was unable to regenerate. Later on the larger mass of this particular specimen approached a leaf of *Posidonia* nearly half an inch from the bottom, and after sending out numerous pseudopodia gradually lifted the body to the edge of the leaf and settled there.

Numerous specimens were removed from their tests by cutting away the chitinous layer at the base and taking the mass of protoplasm out of the test. Such specimens recovered rapidly and sent out pseudopodia like normal specimens in their test. The rate of movement in these specimens was recorded in several cases. There seems to be a rhythmic motion when the specimen is moving in a definite direction. After an interval of rapid motion the whole gradually slows up and then again becomes rapid, although the alternation is somewhat irregular. The following measurements of the time of traveling one-sixth of a millimeter are for consecutive intervals during the time that the movement covered 4 millimeters. It represents the acceleration of speed during a portion of one of these rhythmic intervals:

Minutes.	Seconds.	Minutes.	Seconds.	Minutes.	Seconds.
1	17 20	1	40 30		$\frac{24}{19}$
1 1	11 5	1 1	58 20		$\begin{array}{c} 12\\ 14 \end{array}$
	54 8 13		$54\\50\\40$		15 15 12
1	32		26		14

When two or more specimens are taken from the test and placed near each other pseudopodia are sent out in the usual manner but instead of fusing, as in the case of the parts of a severed specimen, seem to repel one another. The pseudopodia come nearly or quite in contact, then bunch up, forming a considerable mass of greater density, after which the flow is in opposite directions and the protoplasm is withdrawn back to the central masses and movement starts off in other directions. A similar repelling tendency was noted when two species of different families were in motion. In such cases when the pseudopodia came into contact they concentrated in a similar way and the specimens then moved away at a right angle or more from the original line of movement, so as to avoid one another. In all the specimens examined there was no case in which pseudopodia became fused except in the severed parts of the same specimen. Such observations are similar to those observed in Amoebae according to information given me by Professor Schaefer in his work during the same time at the Tortugas. Such observations do not substantiate theories of fusion of various specimens of the same or different species.

Heron-Allen and Earland in their original notes on this species note that the larger specimens as a rule have coarser material in the test and explain the growth of the test as follows: ¹

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With each increase in the size of the test the inclosing wall of the preceding stage is absorbed so as to leave an undivided cavity, the shape of which varies according to the direction and manner in which additions to the original chamber have been made.

In the case of the Tortugas specimens attached to the leaves of *Posidonia* several inches above the bottom it is difficult to see where new material for growth is acquired. The habit which has already been noted, of leaving the test and moving about as a naked mass of protoplasm, may easily account for this change in material. While in this free state new material from the bottom may easily be ingested and carried about until a new attachment is made. Such specimens were placed in a mass of débris composed of fine sand and fine fragments of glass wool, but although some of this material was taken into the body, no new test was formed in any of the specimens kept in confinement.

Some specimens were killed and stained, but only one of these seemed to show a definite nucleus that is shown here (pl. 19, fig. 1). It has a very large nuclear mass with a definite nuclear membrane surrounded by a large mass of ectoplasm, in which are numerous food particles.

The most interesting observations are the rapid motion, the individual character of specimens when in contact, and the power of leaving the test at will.

EXPLANATION OF PLATES.

PLATE 19.

- FIG. 1. Animal of *Iridia diaphana*, test dissolved away, leaving the protoplasmic mass with a single large nucleus, nuclear wall, an irregular mass of ectoplasm containing a few symbiotic algae, and food particles, × 100. Specimen killed and fixed in corrosive sublimate and glacial acetic acid, stained in haematoxylin.
 - 2. Portion of test at periphery from below, \times 50. Central mass of protoplasm with very few algae, most of them being near the peripheral portion and in the radial canals leading from the central mass to the periphery of the test. This specimen was quiescent and showed no pseudopodia at the time it was drawn.
 - 3. A branching mass of pseudopodia irregularly bifurcating, mainly composed of clear protoplasm, but with numerous more concentrated masses and a few of the unicellular algae.

Plate 20.

FIG. 1. Elongate flattened mass with numerous pseudpodia traveling toward the direction of the larger end, \times 50.

2-6. Various stages in the progress of the same individual in a thicker, more nearly rounded form.

PLATE 21.

FIG. 1-7. Various stages in the progress of the same individual as shown in the previous plate. Figure 1 in a subspherical mass apparently about to divide. In figures 2 and 3 the mass flattened out somewhat and withdrew the numerous pseudopodia which had been sent out at the left in figure 1. In figure 4 the fine pseudopodia in the rear had been entirely withdrawn, and in figure 5 those in front had been largely drawn in, leaving the mass quiescent for a short period. There is apparently a small nuclear mass in the forward clear part. In figure 6 lines of pseudopodia are again thrust forward, and in figure 7 motion begins again, a tongue of translucent protoplasm being sent forward and a few fine threads being left at the rear as the mass progresses.